

# Instruction sheet

## AFFINIMIP® SPE Chloramphenicol cartridges

## CLEAN-UP PROCEDURE OF CHLORAMPHENICOL

Users should read all instructions before using this kit.

For laboratory use only

AFFINIMIP® SPE Chloramphenicol is developed and manufactured by AFFINISEP

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## Table of contents

1.	INT	RODUCTION	.3
2.	PRI	NCIPLE OF AFFINIMIP® SPE	.3
3.	PRC	DDUCT INFORMATION	.3
4.	PRE	CAUTIONS FOR USE	.3
5.	REC	COMMENDATIONS FOR HPLC ANALYSIS	.3
6.	GE	NERAL INSTRUCTIONS FOR SPE	.4
e	5.1.	Equipments required	4
e	5.2.	Flow rate	4
e	5.3.	Preparation process	4
7.	CLE	AN-UP PROCEDURE OF CHLORAMPHENICOL:	.4
7	7.1.	Preparation of solutions	4
7	<b>'</b> .2.	Preparation of the loading solution for honey	5
7	7.3.	Preparation of the loading solution for urine	5
7	<b>'</b> .4.	Preparation of the loading solution for shrimp	5
7	<i>'</i> .5.	Protocol for clean-up:	5



### Method for Selective Phase Extraction of Chloramphenicol using Molecularly Imprinted Polymers

#### 1. INTRODUCTION

**AFFINIMIP® SPE Chloramphenicol** has been developed to selectively extract Chloramphenicol from complex matrices such as honey products, urine or shrimp.

By using AFFINIMIP® SPE, the expected result is a clean-up and a pre-concentration of the sample at trace level.

#### 2. **PRINCIPLE OF AFFINIMIP® SPE**

**AFFINIMIP® SPE** is a solid phase obtained by a polymerisation process to create a threedimensional network that recognizes the shape and functional group positions of a template molecule. The **AFFINIMIP® SPE** selectivity comes from the technology of molecularly imprinted polymer (MIP) used during the synthesis.

#### 3. PRODUCT INFORMATION

#### Description of the kit

Each solid phase extraction (SPE) cartridge **AFFINIMIP® SPE Chloramphenicol** of this kit contains 50mg of sorbent in a 1mL cartridge.

#### Information and storage

Storage: Room temperature. Each cartridge has a single use.

#### 4. PRECAUTIONS FOR USE

SPE methods developed for C18 or other sorbents are not appropriate for AFFINIMIP® SPE Chloramphenicol. The extraction procedure described below has been optimized for the extraction of Chloramphenicol from honey. For the treatment of another kind of matrix, please contact us to adapt the extraction procedure.

#### 5. **RECOMMENDATIONS FOR HPLC ANALYSIS**

For HPLC analysis, the following conditions have been used: Column: C18 (USP L1), 2.1 x 50mm, spherical silica gel (Type A), particle size: 2.5µm Mobile phase: Water-acetate ammonium / Methanol



Flow rate: 0.2mL/min

Detection of Chloramphenicol: Mass spectrometer.

This product is also suitable for a clean-up before GC/MS/MS and LC/MS/MS analysis.

#### 6. GENERAL INSTRUCTIONS FOR SPE

#### 6.1. Equipments required

In addition to standard laboratory materials, the following equipments are required for the use of AFFINIMIP® SPE cartridges:

- SPE vacuum manifold
- Nitrogen Mini-vap evaporator or centrifugal concentrator to dry the collected samples

#### 6.2. <u>Flow rate</u>

It is very important to follow the flow rate given in the protocol.

Most especially for the loading, if the sample flow rate is too high, components may not interact sufficiently with the sorbent and the analyte recovery yield will be lower.

#### 6.3. <u>Preparation process</u>

For the preparation of the MIP, a template is required. Chloramphenicol analogues were used instead of Chloramphenicol to prevent false positive signals in case of bleeding.

#### 7. CLEAN-UP PROCEDURE OF CHLORAMPHENICOL:

#### 7.1. <u>Preparation of solutions</u>

- Solution Water – 0.5% Acetic Acid

In a 10mL-volumetric flask, add 50µL of pure Acetic Acid and complete with deionized water.

- Solution (deionized Water - 0.5% Acetic Acid) /Acetonitrile (95/5, v/v)

In a 10mL volumetric flask, add 0.5mL of Acetonitrile and complete with a solution Water-0.5% Acetic acid.

- Solution 1% Ammonia in water (v/v) In a 50mL-volumetric flask, add 1.45mL of Ammonia (35%) and complete with ultrapure water.

- Solution (1% Ammonia in water) /Acetonitrile (80/20, v/v)

In a 20mL volumetric flask, add 4mL of Acetonitrile and complete with the solution 1% Ammonia in water.



#### 7.2. <u>Preparation of the loading solution for honey</u>

5g of honey is diluted with 5mL of deionized water. This solution was mixed under magnetic stirring during 10 minutes and used as the loading solution.

#### 7.3. <u>Preparation of the loading solution for urine</u>

10 mL of urine were adjusted at pH 7 with Ammonia 1%. This solution was mixed and used as the loading solution.

#### 7.4. <u>Preparation of the loading solution for shrimp</u>

5g peeled shrimp were homogenized 2min with a vortex in 20mL of ethyl acetate. Then the solution was filtered on filter paper (25µm). The supernatant was evaporated to dryness and reconstituted in 10mL of Water to obtain the loading solution.

#### 7.5. <u>Protocol for clean-up:</u>

Step (Flow rate)	AFFINIMIP® SPE Chloramphenicol (50mg/1mL)		
Equilibration with	<ul> <li>2mL Acetonitrile</li> </ul>		
(2 drops/s)	<ul> <li>2mL deionized Water</li> </ul>		
	<ul> <li>Do not allow the cartridge to dry during conditioning</li> </ul>		
Loading (L)	1mL of the loading solution		
(1 drop every 2s)			
Washing of interferents	1mL deionized Water		
(1 drop/s)	1mL of (deionized Water-0.5% Acetic Acid)		
	/Acetonitrile (95/5, $v/v$ )		
	• 2mL of 1% Ammonia in water		
	• 2mL of (1% Ammonia in water) /Acetonitrile (80/20,		
	∨/∨)		
Drying :	Force the water down into the cartridge and out the bottom (For this step, you can apply vacuum 1 minutes)		
Washing of interferents	• 250µL diethyl Ether and apply vacuum during 10		
(1 drop/s)	seconds		
Elution (E)	• 2mL Methanol and apply vacuum during 10 seconds		
(1 drop/s)			



The elution (E) is evaporated until dryness under nitrogen with a mini-vap evaporator (or a centrifugal concentrator). The residue is dissolved in mobile phase for further analysis.



## **PRODUCTS LIST**

AFFINIMIP <sup>®</sup> SPE Products	Designation	Description
Multimyco10	AFFINIMIP <sup>®</sup> SPE Multimyco10	selective SPE cartridges 3mL for ZON, OTA, HT-2, T-2, Aflatoxins and Fumonisins
Zearalenone	AFFINIMIP <sup>®</sup> SPE Zearalenone	selective SPE cartridges 3mL for ZON
Ochratoxin A	AFFINIMIP <sup>®</sup> SPE Ochratoxin A	selective SPE cartridges 3mL for OTA
	AFFINIMIP <sup>®</sup> SPE Patulin	selective SPE cartridges for Patulin
Patulin	AFFINIMIP <sup>®</sup> SPE Patulin & Pectinase kit	kit of selective SPE cartridges for Patulin + 50mL pectinase enzyme solution
Deoxynivalenol	AFFINIMIP <sup>®</sup> SPE Deoxynivalenol	selective SPE cartridges 6mL for DON
Phenolics	AFFINIMIP <sup>®</sup> SPE Phenolics	selective SPE cartridges for Phenolic compounds
Estrogens	AFFINIMIP <sup>®</sup> SPE Estrogens	selective SPE cartridges for Estrogens
Zeranol Residues	AFFINIMIP <sup>®</sup> SPE Zeranol Residues	selective SPE cartridges for Zeranol Residues
Bisphenol A	AFFINIMIP <sup>®</sup> SPE Bisphenol A	selective (PP or Glass) SPE cartridges for Bisphenol A
FumoZON	AFFINIMIP <sup>®</sup> SPE FumoZON	selective SPE cartridges for Fumonisins and Zearalenone
Chloramphenicol	AFFINIMIP <sup>®</sup> SPE Chloramphenicol	selective SPE cartridges for Chloramphenicol
Tamoxifen	AFFINIMIP <sup>®</sup> SPE Tamoxifen	selective SPE cartridges for Tamoxifen
Catecholamines	AFFINIMIP <sup>®</sup> SPE Catecholamines	selective SPE cartridges for Catecholamines
Catecnolamines	AFFINIMIP <sup>®</sup> SPE Catecholamines	selective SPE cartridges for Catecholamines
Metanephrines	AFFINIMIP <sup>®</sup> SPE Metanephrines	selective SPE cartridges for Metanephrines
Amphetamines	AFFINIMIP <sup>®</sup> SPE Amphetamines	selective SPE cartridges for Amphetamines
PECTINASE	Pectinase solution	50 mL pectinase enzyme solution
AttractSPE <sup>™</sup> Products	Designation	Description
w/o	AttractSPE <sup>™</sup> W/O	HLB SPE cartridges sorbent
SCX	AttractSPE <sup>™</sup> SCX	Strong Cation Exchange SPE cartridges sorbent
WCX	AttractSPE <sup>™</sup> WCX	Weak Cation Exchange SPE cartridges sorbent
SAX	AttractSPE <sup>™</sup> SAX	Strong Anion Exchange SPE cartridges sorbent
WAX	AttractSPE <sup>™</sup> WAX	Weak Anion Exchange SPE cartridges sorbent
DVB	AttractSPE™ DVB	Reversed Phase Copolymer SPE cartridges sorbent
Anionic & Cationic AttractSPE polymeric cartridges	AttractSPE™ KIT	Kit of 10 cartridges of each sorbent (SAX, WAX, WCX, SCX)

#### For more information:

For more information on our products & services, please visit <u>www.polyintell.com</u>